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REPORT OF DR. WELCH'S REMARKS AND EXHIBITION OF SPECIMENS OF ANIMAL PARASITES AT THE MEETING OF THE JOHNS HOPKINS HOSPITAL MEDICAL SOCIETY, ON MARCH 17th, 1890.

Dr. William H. Welch exhibited gross and microscopical specimens of entozoa which he had found in domestic animals in Baltimore. These parasites had been collected during the last two years as opportunity offered, no attempt having been made to have the collection complete. Dr. Welch said that the interest in animal parasites had been overshadowed of late years by the study of the pathogenic bacteria, but nevertheless the entozoa are of great interest and importance and deserve our careful attention.

The following is a list of the parasites exhibited, all of which were found in Baltimore:

1. FROM THE HORSE:

Spiroptera Megastoma (or Filaria Megastoma). Stomachs were exhibited showing the honey-combed submucous tumors, with ulcerated mucosa, containing the large-mouthed maw-worms. This parasite is not very rare in the horses of this region, but in none of the cases in which it was found by Dr. Welch was there evidence that it had materially damaged the usefulness of the animal.

Strongylus Armatus. Several specimens, gross and microscopical, of verminous aneurism of the anterior mesenteric artery were shown. The interesting histological changes caused by the presence of the palisade worms in the blood-vessels were explained and illustrated by microscopical sections. This parasite with the resulting aneurisms is common in the horses of this region. In one instance it had caused fatal colic.

Gastrophilus Equi. A stomach, the mucous membrane of which near the cardia was completely covered with bots, was presented.

2. FROM THE COW:

Actinomyces Bovis. Although this is a vegetable parasite, the opportunity was taken to show a number of specimens of actinomycotic tumors from the jaws and other parts of the cow. On account of the absence of any satisfactory laws regulating the inspection of méat in this State, the number of cattle affected with actinomyces brought to Baltimore and slaughtered here for the market is unusually large and there is rarely any difficulty in procuring specimens of this disease. Attempts had been made to cultivate the organisms according to the directions given by Boström but without success.



No instance in which the actinomyces had been found in human beings in Baltimore has been recorded.

Cysticercus Taeniae Mediocanellatae—found once in heart-muscle of cow. Strongylus Micrurus. A number of cases of verminous bronchitis and pneumonia in calves due to this parasite had been observed and microscopical sections of lungs so affected were shown.

Filaria Labiato-papillosa (Alessandrini). This worm was found free on the peritoneal surface of a cow without causing any apparent injury.

3. FROM THE SHEEP:

Taenia Expansa-found in the small intestine. Common.

Strongylus Contortus—found in large numbers in the stomach of several sheep which had died at Druid Hill Park without other apparent cause of death.

4. FROM THE HOG:

Echinococcus. Echinococci were found in several hogs which had been born and reared in the neighborhood of Baltimore, but this parasite is not common in this region. Sections of liver were shown illustrating every stage of development of this bladder-worm from cysts smaller than a pea up to cysts as large as an orange. The taenia echinococcus produced by feeding the cystic worms to a dog was shown.

Cysticereus Cellulosae. The pork-measle had been found in only a few instances.

Echinorhynchus Gigas. This worm is common in the swine of this region and appears to be common throughout the United States. The ulcerated and necrotic patches in the inner wall of the small intestine caused by the attachment of the thorn-headed worm, bear some resemblance to the necrotic foci resulting from hog cholera. In only one instance had the gut been actually perforated by this worm.

Trico-cephalus Crenatus—very common in the coecum.

Ascaris Suilla, believed by many to be identical with the ascaris lumbricoides, was not very often met with. In one case the small intestine for a distance of 40 cm. was found packed full of ascarides, but there were no evidences of intestinal obstruction and it is questionable whether this accumulation of the worms, so as apparently to obstruct the gut, is not a post-mortem occurrence.

Sclerostoma Pinguicola (Verrill), believed to be identical with Stephanurus Dentatus (Diesing), although this identity is not absolutely certain. This interesting worm was found in the abdominal adipose tissue and in the liver. As the presence and mode of migration of this parasite in the liver of swine have not hitherto been described, Dr. Welch showed a number of specimens making these points clear. The sclerostoma pinguicola was found in the livers of eleven swine, and, while not very frequent, cannot be considered a rarity in this situation. It is found, often in large numbers, in the main trunk and branches of the portal vein, which then usually contain parietal or occluding thrombi in which the worms are imbedded. The real habitat of the worm is, however, the connective tissue around the portal vessels. In this tissue it burrows its way, producing inflammatory masses of new connective tissue rich in leucocytes. section of the liver, nodules and bands with sinuous cavities containing a brownish or reddish white purulent fluid are observed. Similar nodules are also visible projecting on the surface of the liver. In these sinuous cavities the parasite may be found, or it may be absent, having made its way to other parts. The worm finds its way into the portal vessels by ulceration from the peri-portal tissues through the walls of the vessels, and in several instances the worm could be demonstrated partly within and partly without the vein. Before actual penetration into the vein there occurs a bulging inward of the vessel wall on which forms a thrombus composed primarily of blood plates. One of the most interesting histological changes produced by the invasion of this parasite into the liver is an extensive new growth of the mucous glands in the walls of the bile ducts. This new growth occurs in the areas of newly formed connective tissue in the neighborhood of the parasites. Actual adenomatous formations of undoubted parasitic origin are produced in this way. The same alteration has been recently described and pictured by Schaper in connection with distomatosis of the liver (Deutsche Zeitschrift für Thiermedecin, Bd. xvi, p. 1). On sections of the worms, as found in the liver, leucocytes with wellstained nuclei, resembling those in the fluid of the cavities containing the worms, can be seen in the intestinal canal of the worm, but whether this can be interpreted as evidence that the pus cells produced by the presence of the worm actually serve as its food is uncertain. Sometimes the lesions of the liver, which have been described, were extensive, the surface and interior of the organ presenting a large number of parasitic foci, and it would seem as if such an extent of the disease must be injurious, but no cases were observed in which the death of the animal could be attributed to the presence of the sclerostoma pinguicola. The identity of the parasite as found in the liver and as found in the well known foci in the abdominal fat tissue was demonstrated. Instances in which the main trunk of the portal vein as it enters the liver was completely occluded by a thrombus containing the sclerostomata were exhibited.

Strongylus Paradoxus. This strongyle is extremely frequent in the bronchi of swine in this region. Its presence was found to be the direct cause of death in three pigs. In these cases the trachea and bronchi contained an almost incredible number of strongyles, so that on sections of the lung, the medium-sized and small bronchi appeared filled with worms. The strongylus paradoxus is usually associated with some bronchitis and often with broncho-pneumonia, but it may be present, even in large numbers, without either bronchitis or pneumonia. In fact in one of the fatal cases mentioned in which the

strongyles appeared to fill the medium-sized and small bronchi and were present in enormous numbers in the trachea and large bronchi there was no trace of pneumonia and scarcely any bronchitis. The animals suffered from extreme dyspnoea and the heart pulsated so violently as to be visible at a distance and to communicate its motion to the entire thorax. At the autopsy the right ventricle was hypertrophied. The favorite habitat of the worms when present only in moderate number is in the bronchi in the posterior part of the caudal lobes, and sometimes their number is so small that considerable searching is required to detect them. There is usually a little muco-pus in the bronchi where the strongyles are lodged even when there is no general bronchitis. The adjacent lung parenchyma is often emphysematous or on the other hand it may be simply atelectatic or it may be the seat of a broncho-pneumonia. Frequently there is broncho-pneumonia of the ventral lobes when the worms are to be found only in the posterior parts of the caudal lobes. The broncho-pneumonia associated with the strongylus paradoxus appears as a brownish or grayish red consolidation in which the individual lobules and lobulettes can be felt and seen as nodules. The affected part is not much swollen and there is generally no pleuritic exudation. On microscopical section the air cells contain leucocytes. epithelioid cells, sometimes fibrin and red blood-corpuscles and frequently ova of the strongyles. These ova are often partly or completely enclosed within giant cells. By a combination of Weigert's fibrin stain and picrocarmine very beautiful pictures in which the ova are stained blue and the cells red and yellow with picrocarmine can be obtained. A very instructive lesson in embryology is furnished by the ease with which all stages of development of the ova from the simple cell up to the developed embryo worm can be followed in these preparations.

5. From the Dog:

Taenia Cucumerina. This is by far the most common tapeworm of the dogs used for experiment in the laboratory, being found in 60 per cent, of these animals,

Taenia Serrata. Specimens produced by feeding dogs the cysticerci pisiformes from the rabbit were shown.

Taenia Echinococcus was found only in dogs artificially fed with the echinococcus from the hog.

Eustrongylus Gigas. A specimen of this worm, 95 cm. long, was found free in the peritoneal cavity of a dog and was exhibited. It had been found three times in this situation in dogs used for experiment in the laboratory.

Strongylus or Dochmius Trigonocephalus (Uncinaria Trigonocephala) was found in the small intestine of 70 per cent. of the dogs used in the laboratory, a much larger proportion than has hitherto been observed. The head of the worm was often found imbedded in the mucous membrane and surrounded by a small extravasation of blood.

Sometimes scanty, it was in many cases found in large numbers. Positive evidence that the parasite was the cause of anemia in the animals could not be found. It was occasionally met with in the stomach as well as in the small intestine.

Tricocephalus Depressiusculus appears to be a constant inhabitant of the dog's coecum.

Ascaris Marginata (doubtless a variety of Ascaris mystax) was found in only a few cases. It was found both in the stomach and small intestine.

The *filaria immitis* was not met with in any of the dogs used in the pathological laboratory and is not common in this region. It, however, occurs here as it has been observed occasionally in the Biological Laboratory of the Johns Hopkins University.

6. FROM THE RABBIT:

Coccidium Oviforme was present in the liver of one-third of the rabbits used for experiment in the laboratory. Coccidia are also common in the intestines of rabbits. Attention was called to the existence of small, opaque, grayish-white patches, suggesting somewhat a superficial necrosis, on the surface of the mucous membrane of the small and the large intestine. These patches contain large numbers of coccidia, often enclosed in epithelial cells.

Cysticercus Pisiformis is common in the peritoneal cavity. Specimens were shown illustrating the nodules and scars caused by the migration of this parasite through the rabbit's liver.

From the rat and cat were shown respectively the cysticercus fusciolaris in the liver and the taenia crassicollis in the intestine.

No instance of distoma in the animals examined was discovered and in general distomatosis of domestic animals appears to be rare in this region. Through the kindness of Prof. Brooks, Dr. Welch was in the possession of a number of living water snails, many of which were infested with cercaria and living specimens of these interesting forms in the life history of distoma were exhibited under the microscope. These snails were obtained in the neighborhood of Baltimore, so that opportunity for infection with distomata is present here.

REPORT OF DR. WELCH'S REMARKS ON THE DIPLOCOCCUS PNEUMONIAE AT THE MEETING OF THE JOHNS HOPKINS HOSPITAL MEDICAL SOCIETY, ON FEBRUARY 17th, 1890.

Dr. William H. Welch reported results of his bacteriological examinations of cases of acute croupous pneumonia, and exhibited cultures and microscopical specimens of the micro-organisms found in these cases. In all of the cases examined, ten in number, the Diplococcus Pneumoniæ was found and isolated in pure culture. This organism is probably constantly present in croupous pneumonia, and the evidence that it is the specific cause of this disease is very strong. In the cases examined by Dr. Welch, roll cultures or plate cultures were made from the affected parts of the lungs, from the spleen, from complicating lesions and frequently from the blood and other situations. Nutrient agar, glycerine agar and gelatine agar, according to Guarnieri's formula, were the media usually employed for this purpose. The growth of the pneumo-coccus on the last named medium is particularly luxuriant. In addition mice and rabbits were inoculated with bits of hepatized lung, with the spleen, etc. In one instance the presence of the pneumococcus would have been overlooked in consequence of the prevalence of other organisms, if the precaution had not been taken to inoculate animals with the tissues. Dr. Welch expressed the opinion that the diplococcus pneumoniæ could not be said to be absent simply on the ground of negative results from cultures. This result must be controlled by the inoculation of susceptible animals. Moreover, even the failure to kill mice and rabbits by inoculation can not be considered conclusive evidence of the absence of the diplococcus pneumoniæ, for this organism may be present in the human body in cases of croupous pneumonia in a form incapable of killing rabbits or even mice.

One of the most interesting and extraordinary properties of this organism is the variation in its virulence when tested upon animals. All who have experimented with cultures of the diplococcus pneumoniae have called attention to its rapid loss of virulence when cultivated in artificial media. It is important to note that a progressive diminution of virulence may occur likewise in the lungs and other organs where the pneumo-coccus is present in cases of croupous pneumonia. In one case of croupous pneumonia dying in the stage of grey hepatization Dr. Welch found very numerous colonies of the diplococcus pneumoniae, mixed with a few colonies of the staphylococcus aureus in agar plate cultures from the consolidated lung. A rabbit inoculated with a piece of the lung died on the fifth day with extensive local reaction and large spleen. The pneumo-cocci were abundant near the seat of inoculation, and were found in considerable number, although fewer than usual, in the blood. Plate cultures (agar) made from the swollen spleen

(after burning thoroughly the surface) of this patient showed in very large number, colonies of the pneumo-coccus. No other species of organism was present in these cultures from the spleen. The same pneumo-coccus was likewise observed in cover-glass preparations from the fresh spleen. The organism isolated from the spleen conformed in every particular as regards its morphology, its behavior in artificial culture media, its short vitality, its color-reactions to the pneumo-coccus found in the lung, the so-called Fraenkel-Weichselbaum pneumo-coccus. Nevertheless it was devoid of its customary virulence, for a mouse and a rabbit inoculated subcutaneously with a bit of the spleen survived, and mice and rabbits inoculated with pure cultures of the organism likewise survived, the only reaction being a small abscess or purulent infiltration of the tissues at the point of inoculation. In this case, therefore, the diplococcus pneumoniæ was present in large numbers in the hepatized lung in a weakened state as regards its virulence, and in the spleen in a condition incapable of killing rabbits and even mice. Every grade of virulence was observed in the pneumo-cocci found in the different cases, from the degree just described as incapable of killing mice and rabbits up to the degree in which rabbits were killed in 36 to 48 hours by an acute septicæmia. In two cases the pneumo-coccus did not kill rabbits but it did kill mice, so that the latter animals are more susceptible and are to be preferred for inoculating tissues in cases of croupous pneumonia. In three cases rabbits survived the inoculation for more than five days, the longest duration being twelve days. Inoculation of rabbits with exudation present in the bronchi or in the trachea sometimes caused a rapidly fatal sputum-septicæmia even when the pneumo-coccus present in the hepatized lung showed weakened virulence. In general the impression was obtained that the most virulent forms of pneumo-cocci are to be found in the sputum, in the freshly hepatized lung or at the margin of an advancing pneumonia, whereas the pneumo-cocci present in advanced stages of hepatization and in the spleen are likely to be less virulent. This progressive diminution in virulence, as tested upon animals, of the pneumo-coccus in the consolidated lung and other organs in cases of croupous pneumonia is surely a significant circumstance. In no instance had Dr. Welch as yet been able to isolate the pneumo-coccus in a state capable of producing pneumonia in dogs, although not less than 5 cc. of a bouillon culture 24 hours old (in the thermostat at 37° C.) of the pneumo-coccus, obtained from a rabbit dead 40 hours after inoculation, had been injected directly into the lung. Other observers, however, had obtained positive results by this method of inoculation. Others have found pneumo-cocci producing more rapidly fatal septicæmia in rabbits than resulted from any of the cultures obtained from the cases examined by Dr. Welch.

In six of the ten cases the diplococcus pneumoniæ was found in pure culture in the hepatized lung as determined by microscopical examination and by plate or roll agar cultures; in two cases it was present in combination with the staphylococcus pyogenes aureus; in one case, in addition to the pus staphylococci, the streptococcus pyogenes was present in considerable number; and in one case with gangrenous foci there were present, besides the diplococcus pneumoniæ and the pus staphylococci, a short oval bacillus and a long

slender bacillus not developing in plate cultures (aerobic cultures). In one of the cases in which the staphylococcus pyogenes aureus was present, several small abscesses had formed in the hepatized lung. In four cases the diplococcus pneumonia was found in pure culture in the spleen, which in all of these instances was swollen and soft; in one case it was isolated from the kidney and in one from the heart's blood. In all instances (five) in which the pleuritic exudate accompanying acute croupous pneumonia was examined by culture methods, the diplococcus pneumonia was found in this exudate. In three cases of empyaema following acute croupous pneumonia (these do not belong to the ten cases now under consideration) the diplococcus pneumonia was found; in two in pure culture, in one in combination with staphylococcus pyogenes aureus and albus. The pneumo-bacillus of Friedländer was not found in any case.

Dr. Welch described the morphology and behavior in culture media of the diplococcus pneumoniæ. He had found that a convenient way of staining the capsules was to treat the cover glass specimens, dried and treated in the usual way, with glacial acetic acid and then without washing off the acid to drop on the cover glass aniline-oil gentian-violet, which can be allowed to drain off and can be renewed two or three times. Attention was called to broad pale rods corresponding to empty capsules sometimes found in the fluid of empyaema following pneumonia. These may be 10–30 mm. long and 2–3 mm. broad. They stain faintly, may be entirely empty or may contain one or more deeply stained pneumo-cocci. They correspond to the capsular substance around rows of pneumo-cocci, the cocci having partly or wholly disappeared.

For staining sections, Weigert's fibrin stain had given most excellent results. A method which had yielded good and quick results was to dry thin frozen sections from the fresh organs upon the slide and then to heat them for half an hour at a temperature of 110° to 115° C. These can then be stained like cover-glass specimens. This method is often employed in the laboratory for staining fresh tissues in general for micro-organisms.

The statements of Fraenkel and others were confirmed as to the behavior of the diplococcus pneumoniæ in artificial culture media, its susceptibility to slight changes in the reaction and composition of the medium, its brief vitality, etc. As already mentioned, Guarnieri's gelatine-agar medium was found to be particularly suitable.

The history of our knowledge of this organism from its discovery by Sternberg in September, 1880, up to the present time and the results and conclusions reached by the various investigators of its relation to croupous pneumonia were briefly reviewed. The frequent presence of the pneumococcus in the saliva of healthy persons is upon the whole an assistance to us in the explanation of the various factors concerned in the causation of croupous pneumonia.